## **Nematodes: Lords of the Flies?**

Biting flies that pester cattle could soon get a taste of their own medicine. Agricultural Research Service (ARS) and University of Nebraska-Lincoln (UN) scientists are testing a way to fight the flies in feedlots where they gather and breed. They're using tiny parasitic roundworms called nematodes that prey on the flies' maggot offspring.

Exploring new, nonchemical ways of protecting cattle is the objective of a 3-year-old project by entomologist David B. Taylor at ARS' Midwestern Insect Livestock Research Unit in Lincoln. In nematodes he sees a biocontrol agent that could be part of an integrated fly-control program at the feedlot along with traps, manure management, sanitation measures, and parasitic wasps.

Since 1999, Taylor and UN associate Thomas Powers have screened 20 species and 50 strains of fly-infecting nematodes for their abilities. Of particular interest were those capable of persisting in cow manure long enough to kill house and stable fly larvae over an entire season.

In Nebraska, where beef cattle are the top agricultural commodity with annual sales of \$5.5 billion, stable flies are considered even worse pests than house flies. That's because attacks by swarms of these relentless biting flies cause blood loss, stress, and feed-efficiency problems. The flies may also harbor disease organisms, and they cost the U.S. beef and dairy cattle industry up to \$1 billion in annual production losses.

Taylor's and Power's strategy calls for battling the pests in manure around feedlots or in soiled calf pen bedding. That's where 80 percent of the flies' brood hatch and feed. And prolific breeders they are—hundreds of maggot offspring emerge from a single pound of manure. Therein lies the nematodes' appeal, for a mating pair of these roundworms can produce 5,000 to 10,000 offspring in a single maggot in less than 2 weeks. Says Taylor, "The nematodes actually reproduce faster than the flies."

In experiments, up to 99 percent of fly maggots died within 48 hours of infection by the top fly fighter, *Steinernema feltiae*. In the laboratory, "the nematodes can live in bovine manure for 4 to 6 weeks without hosts," says Taylor. In feedlots, he adds, "We'd like to apply them in May and get season-long fly control." Chemical insecticides, in contrast, must be reapplied, and flies can develop resistance to them.

The researchers are testing ways to apply the nematodes on manure and protect them from drying and ultraviolet light. About 1 million nematodes per square meter are used, but lower rates might suffice, keeping the costs closer to chemical controls. If the approach works, the nematodes could also be used to fight corn rootworms in manure-fertilized fields, says Taylor.—By **Jan Suszkiw**, ARS.

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## **Leafminers Identified Faster**

Two leafmining flies—*Liriomyza huidobrensis* and *L. langei*—are pests of many vegetable and flower crops, including peas, beans, melons, onions, tomatoes, potatoes, celery, garlic, lettuce, chrysanthemums, and carnations. Their larvae tunnel inside leaves and other plant parts as they feed, leaving winding trails called mines that are visible through the leaf surface.

During outbreaks, these insects can cause severe damage, resulting in substantial economic losses. Originally present only in the western United States and South America, they have now invaded Europe, the Middle East, and Asia.

L. huidobrensis and L. langei are so similar in form and structure that they were once considered to be one species. But ARS molecular biologist Sonja Scheffer and biological sciences technician Matthew Lewis used DNA sequence data from mitochondrial and nuclear genes to show that there are actually two species. DNA data also showed that the invasive leafminers causing extensive crop damage around the world are L. huidobrensis, not L. langei. Currently, the highly invasive L. huidobrensis is not known to be present in the United States.

Using DNA sequence data is an expensive, time-consuming way to identify species. Now Scheffer has refined a less expensive, faster molecular method to differentiate the two pests. It uses polymerase chain reaction combined with restriction fragment length polymorphism (PCR-RFLP) analysis. This method can be used with adult, larval, or pupal leafminer specimens. It was developed to provide researchers, pest managers, and quarantine officers with a simple and quick molecular method to differentiate *L. langei* from *L. huidobrensis*.

In early 2000, Scheffer confirmed the accuracy of PCR-RFLP by applying it to 31 fly specimens for which DNA sequence data had clearly identified the species. She tested 52 more specimens from recently introduced leafminer populations in Sri Lanka, South Africa, and Canada and found all to be *L. huidobrensis*.

PCR-RFLP can be performed by anyone with access to a laboratory that has a PCR thermocycler and associated paraphernalia—an increasingly common piece of equipment.

"The entire set of procedures—from DNA extraction to final identification—can be completed within a single working day," says Scheffer, "compared to several days and additional expense for DNA sequencing."

But there's more opportunity for misidentification with this method than with sequence data, because PCR-RFLP banding patterns may be shared by multiple species. For this reason, it is appropriate to use a particular PCR-RFLP test only with those species for which the test was developed.—By **Jennifer Arnold**, formerly with ARS.

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